# Teacher packs in Experimental Science

## BIO Pack 1

# Analysis of the effect of saliva against starch

***Pack contents:***

A. Teacher's Guide
B. Students Guide
C. Assessment – Student's sheet
D. Extension to experiments
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F. Health and Safety
G. Evaluation pack

***Curriculum areas covered:*** Food and Nutrition in Plants and Animals

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**Title: Analysis of the effect of saliva against starch**
***Target group:*** Colleges of Education
***Also suitable for:*** Junior High Schools

***Learning outcomes:***

These are the learning outcomes expected after students have gone through this Pack

1. Knowledge and understanding

   KN1 Demonstrate knowledge and understanding of the action of enzymes

2. Cognitive skills

   Be able to:

   CS1 describe, analyze and interpret scientific data
   CS2 apply knowledge and understanding of this experiment to suggest additional lines of investigation

3. Key Skills

   KS2 Be able to process and present data

4. Practical skills

   PS1 Be able to make and record observations and measurements and report results


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# A. **Teacher's Guide**

### Sample Assessment Questions

Assessment questions should be linked to the learning outcomes specified at the beginning of the investigation (See Section 4). A few are suggested below.

1. Which other organ in your body produces amylase?
2. Can you think of another way of following the progress of the reaction?
3. Suppose you obtained the original iodine colour just after 30 seconds how could you modify the procedure to enable you observe the progressive hydrolysis of starch?
4. If you did not see any colour change at all, what could be the possible explanation?

Note:

The procedure as described in the instructions for students is rather simplified and approximate. Depending on the Level of the students and the availability of equipment, more precise volume measurements may be used to refine the data on the relationship between the rate of reaction and relative concentrations of substrate and enzyme.

If possible, the effect of changes in temperature and pH on the rate of reaction can also be investigated. For instance students could work out the optimal temperature for the enzyme and relate it to the normal temperature of the human body

On the other hand, where it is difficult to obtain even the simple equipment listed, suitable substitutes may be used, provided they enable the student obtain comparable results or lead them to the desired learning outcomes.

## B. **Student Guide**

**Purpose:**

To test the hypothesis that saliva contains an enzyme that hydrolyses starch.

**Background to Experiment**

Physiological activities in the mammalian body involve the activity of several enzymes. These include: catalase, amylase, lipase, pepsin, and trypsin. Find out from your textbook what role each of these enzymes plays in the mammalian body.

As an enzyme works, it combines with its substrate and converts it to product(s). In this activity, you will identify enzyme activity by observing changes in the amounts of substrate and product(s).

| | ENZYME | |
|---|---|---|
| SUBSTRATE | - - - - - - - - > | PRODUCTS |

As you carry out the activity, it is important for you to understand what you are doing, why you are doing it in the manner specified, and what the results mean.

In this exercise, you will investigate the presence and activity of salivary amylase, an enzyme which digests starch in our mouths.

Salivary amylase is produced by the salivary glands. If amylase is added to a solution of starch, it is expected to catalyse the hydrolysis (digestion) of starch (a polysaccharide) to form maltose (a disaccharide).

| **SUBSTRATE** | **ENZYME** | **PRODUCTS** |
|---|---|---|
| | Amylase | |
| starch | -------- - - - - - - - - - - > | maltose + maltose + -- etc. -- |



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## Iodine Test for Starch

The starch test consists of adding a drop of iodine solution ($I_2KI$) to the sample to be tested. If the iodine retains its yellow-brown color, starch is absent. If a purple or blue- black color forms, starch is present, and the deeper the color, the greater the amount of starch. Thus the progress of the reaction can be visualized by testing the reaction mixture for the disappearance of the substrate (starch), using the iodine test.

## Equipment/ Materials

Starch (extract or raw tuber e.g. yam, cassava, arrowroot)
Cling film
Cup or glass
Detergent (for cleaning glassware)
Pen/ marker with indelible ink (for labeling various containers or droppers)
White plate/ tile
Tablespoon
Stop-watch or clock with seconds hand or phone with 'stop-clock' function
Paper tissues
Iodine/ potassium iodide
Pipettes or medicine droppers
Glass stirring rod
Beakers
Spatula
Test-tube rack

## Other requirements

- The location chosen for the experiment should be well illuminated.
- It is necessary to avoid eating (for at least 15 minutes) or smoking (for at least one hour) before carrying out the experiment.
- You need a notebook and a pen for recording your results.



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**Experimental Procedure**

Wash all glassware or other containers thoroughly with hot water and detergent

Rinse well in hot clean water and allow to drain.

Next, prepare three solutions as follow

I. Starch solution

Boil some water in a kettle or saucepan
Take a beaker, label it 'starch solution' and put into it one level spatula of starch
Fill a test tube a quarter full of cold water and add this to the starch in the beaker.
Stir until you have a white suspension
Pour hot water into beaker till it is almost full and stir with a glass rod – this should produce a hot, slightly cloudy solution of starch.
Cover the beaker with cling film to keep out the dust and set it aside to cool.

*Note*

Wait till the solution is at room temperature before use. In order to save time, this solution can be prepared a day before (or well in advance on the same day) and stored (in a fridge but allowed to warm up before use) till it is time to be used.

II. Iodine solution

Label a test tube 'iodine' and place in it one level spatula of potassium iodide crystals. Add distilled water till the test tube is half-full. Next, add two small crystals of iodine and shake the mixture until the particles of iodine have dissolved as far as possible. The solution is expected to have the colour of 'tea without milk'. Place the test-tube in the test tube rack.

III. Amylase solution

Measure a tablespoon of cold water into a cup or glass.
Label a second beaker 'enzyme'.
Put the water from the cup into your mouth and chew, swirl or gargle it vigorously for about 15 seconds (use the stop watch or clock). Now spit the solution into the beaker.

NOTE

Label your solutions clearly and put them to one side of your work area. Why is it important to label your solutions?

1. Testing the assay
    a. Have ready the clean, white plate/tile

Label one dropping pipette A and the other B and put a rubber teat on each of the pipettes

Use pipette A to take in some of the iodine solution and put a ring of spots (one or two drops per spot) of the solution around the plate/tile. Store pipette A in the test-tube



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containing the iodine solution in the test-tube rack, in case you need to repeat the experiment.

b. Pour about 1cm depth of starch solution into a clean test-tube. Take dropping pipette B and use it to transfer a drop of the starch solution from the test-tube to an iodine drop on the plate. Ensure that in the process, you do not contaminate the tip of the pipette with iodine solution. View the plate in bright light and note the grainy dark blue (or almost black) spot. This shows that starch is present and what sort of indication you get with the iodine assay.

2. Applying the assay to saliva

a. Now add about a 2cm depth of enzyme solution (i.e. mouth washings) to the starch solution in the test-tube. Shake the mixture and using pipette B suck up a full pipette of the mixture. The enzyme reaction will go on inside the pipette. Squeeze out one drop of the mixture onto successive iodine spots every 30 seconds (use the stop-watch as a guide). As soon as you have added the drop, observe and note any colour change. Continue adding drops. If you do observe a colour change, continue until addition of one drop of reaction mixture to a spot just fails to give a colour change. The total time that has elapsed since the enzyme was mixed with the starch is the achromatic time. Record this time in your Notebook.

b. At the end of the experiment, discard all solutions down the sink and then pour down some household bleach. Also immerse all glassware used in some diluted household bleach solution overnight. Then rinse in clean hot water, allow to dry and store in a suitable place.

***Note: Household bleach may be harmful and should be handled according to the manufacturer's instructions***

3. **Reflection on the experiment**

a. Take some time to reflect on the experiment and ensure that you understand fully the procedure followed. If you need clarification, have a discussion with you teacher or colleagues.
b. Do you think this investigation could be made in a different way?
c. How would you explain the results obtained?



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C. **Assessment – Student's sheet**

On completion of the experiment, you should answer the following questions:

1. Which other organ in your body produces amylase? (KN1)

2. Can you think of another way of following the progress of the reaction? (CS2)

3. Suppose you obtained the original iodine colour just after 30 seconds how could you modify the procedure to enable you observe the progressive hydrolysis of starch? (CS2)



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4. If you did not see any colour change at all, what could be the explanation? (CS2)



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## D. **Extensions to experiment**

The rate of the reaction is expected to vary with changes in temperature of the enzyme and substrate mixture. The effect of temperature could be investigated and it would be interesting to measure the optimal temperature and compare it with the normal body temperature ($37^{o}$C).

## E. **Useful links**

http://www.biotopics.co.uk/nutrition/amylex.html

http://eve.kean.edu/~breid/enzyme/enzyme.html

http://www.eng.umd.edu/~nsw/ench485/lab5.htm

## F. **Health and Safety**

Care required with the use of household bleach (see Student Guide)

## G. **Evaluation**

You may write here your observations on the Pack, challenges encountered in carrying out the activity and suggestions for improving usefulness of the Pack.



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